Detection of protein ubiquitination.

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Public Summary:

Scientific Abstract:
Ubiquitination, the covalent attachment of the polypeptide ubiquitin to target proteins, is a key posttranslational modification carried out by a set of three enzymes. They include ubiquitin-activating enzyme E1, ubiquitin-conjugating enzyme E2, and ubiquitin ligase E3. Unlike to E1 and E2, E3 ubiquitin ligases display substrate specificity. On the other hand, numerous deubiquitylating enzymes have roles in processing polyubiquitinated proteins. Ubiquitination can result in change of protein stability, cellular localization, and biological activity. Mutations of genes involved in the ubiquitination/deubiquitination pathway or altered ubiquitin system function are associated with many different human diseases such as various types of cancer, neurodegeneration, and metabolic disorders. The detection of altered or normal ubiquitination of target proteins may provide a better understanding on the pathogenesis of these diseases. Here, we describe protocols to detect protein ubiquitination in cultured cells in vivo and test tubes in vitro. These protocols are also useful to detect other ubiquitin-like small molecule modification such as sumolyation and neddylation.